EFFECT OF DIETARY CALCIUM AND VITAMIN D ON CORTISOL CONCENTRATIONS IN MORBIDLY OBESE WOMEN

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Abstract

Studies have shown inadequate intake of micronutrients, such as calcium and vitamin D, by obese individuals, and its relationship with cortisol in this population are of great interest among researchers. This study evaluates the relationship between dietary concentrations of calcium and vitamin D and cortisol metabolism in morbidly obese women. The present study involved 58 women divided into a case (29 morbidly obese) and control group (29 eutrophic women). Body mass index and waist circumference were measured, and calcium and vitamin D intakes were analyzed using dietary records of three days and Diet Pro 5i® software (AS Sistemas, Viçosa, Brazil). In addition, serum and urinary cortisol were determined by chemiluminescent methods. Ingestion of calcium and vitamin D by the morbidly obese (761.54 ± 353.21 mg / day and 8.07 ± 9.86 mg / day, respectively) was lower than the recommended values. There was a significant correlation only between vitamin D intake and adiposity parameters (P<0.001). There was no correlation between intake of calcium and vitamin D and serum and urinary cortisol (P>0.05). In this study, obese individuals ate foods low in calcium and vitamin D. In addition, the intake of this vitamin was negatively correlated with adiposity parameters. The morbidly obese had normal and normal cortisol concentrations, and thus, hypercortisolemia and hypercortisoluria were not evidenced.

Keywords:
Morbid obesity; Calcium, dietary; Hydrocortisone; Vitamin D; Diet.

Resumo

Estudos têm demonstrado ingestão inadequada de micronutrientes, como cálcio e vitamina D, por indivíduos obesos, e sua relação com o cortisol nesta população são de grande interesse entre os pesquisadores. Esse estudo avalia a relação entre as concentrações dietéticas de cálcio e vitamina D e o metabolismo do cortisol em mulheres obesas mórbidas. O presente estudo envolveu 58 mulheres divididas em um grupo caso (29 obesos mórbidos) e grupo controle (29 mulheres eutróficas). O índice de massa corporal e a circunferência da cintura foram medidos e o consumo de cálcio e vitamina D foram analisados utilizando-se registros dietéticos de três dias por meio do software Diet Pro 5i® (AS Sistemas, Viçosa, Brasil). Adicionalmente, o cortisol sérico e urinário foram determinados por métodos quimioluminescentes. A ingestão de cálcio e vitamina D por obesos mórbidos (761,54 ± 353,21 mg / dia e 8,07 ± 9,86 mg / dia, respectivamente) foi inferior aos valores recomendados. Houve correlação significativa apenas entre ingestão de vitamina D e parâmetros de adiposidade (P <0,001). Não houve correlação entre ingestão de cálcio e vitamina D e cortisol sérico e urinário (P> 0,05). Neste estudo, indivíduos obesos comeram alimentos com baixo teor de cálcio e vitamina D. Além disso, a ingestão desta vitamina foi negativamente correlacionada com os parâmetros de adiposidade. Os obesos mórbidos apresentaram concentrações normais e normais de cortisol e, portanto, não foram evidenciadas hipercortisolemia e hipercortisolúria.

Palavras-chave:
Obesidade Mórbida; Cálcio, dietético; Cortisol; Vitamina D; Dieta.

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INTRODUCTION

Obesity is a highly prevalent chronic disease that has become a global epidemic and a significant cause of death in recent years. This disease is related to several co-morbidities, such as diabetes, atherosclerosis, dyslipidaemia, hypertension, cardiovascular disease and some cancers[1,2].

Adipose tissue is a multipurpose organ that performs important functions such as energy storage and endocrine action. This tissue produces and releases several molecules called adipokines or adipocytokines and is considered an important endocrine organ that regulates physiological and pathological processes, including immunity and inflammation. The anatomical location of fat deposits influences the production of these molecules; visceral fat particularly is metabolically more active[3,4].

Several studies have been conducted with a view to clarify the involvement of micronutrients in controlling body weight. Recent studies have shown inadequate intake of various nutrients in the obese population, and minerals, calcium in particular, have been of great interest to researchers[5,6]. A study conducted by Sánchez et al.[7] showed reduced levels of calcium in the diet of morbidly obese women. Similarly, Rodríguez-Rodríguez et al.[8] found that calcium intake was inversely related to body mass index.

The exact mechanism by which calcium intake leads to a reduction of abdominal obesity is unclear, but autocrine cortisol production from adipose tissue could explain this effect. It has been shown that calcitriol stimulates expression of the enzyme 11β-hydroxysteroid dehydrogenase type 1 (11β-HSD-1) that catalyses the conversion of cortisone to cortisol, which is involved in fat deposition mainly in the abdominal region. Thus, it is suggested that a calcium-rich diet, which suppresses calcitriol levels, leads to decreased body fat accumulation by reducing the production of cortisol in adipose tissue[9,23].

It is important to understand the close relationship between calcium and vitamin D during the metabolic events of adipogenesis. Vitamin D and its association with reduced body weight has been the subject of several studies[10].Current literature supports a relationship between low serum levels of vitamin D and increased fat percentage and body mass index[1,11]. However, the mechanisms underlying this association have not been fully elucidated.

Although some studies have already shown inadequate intake of micronutrients in the obese population, mechanisms involving dietary calcium and vitamin D intake and their relationship with cortisol are poorly understood. Determination of the content of these micronutrients in the diet, as well as their relationship with serum and urinary cortisol, may help to clarify their influence on cortisol metabolism in obesity. Therefore, this study was conducted in order to evaluate the relationship between dietary calcium and vitamin D intake, and cortisol concentrations in morbidly obese women.

MATERIALS AND METHODS

This cross-sectional study evaluated 58 women aged between 20 and 59 years. The participants were divided into two groups: obese group (morbidly obese, n=29) and control group (normal weight, n=29). Obese women were recruited from the spontaneous demand of a private clinic in Teresina, Piauí, Brazil and the control group consisted of volunteers who attended the Experimental Nutrition Laboratory of the Federal University of Piauí from a public call.

The participants were selected according to the following criteria: body mass index between 18.5 and 24.9 kg/m² (control group) or ≥40.0 kg/m² (obese group); non-smokers; not pregnant or lactating; absence of diabetes mellitus, cardiovascular disease, cancer, chronic renal failure, and liver disease; and not taking vitamin and mineral supplements and/or medicines that could affect cortisol levels.

This study was approved by the Research Ethics Committee of the Federal University of Piaui (protocol number 0460.0.045.000-11) and conducted in accordance with the Declaration of Helsinki. All participants gave their full consent.

Assessment of Nutritional Status

To assess the nutritional status, body mass index was calculated as the body weight divided by the square of the height. Nutritional status was classified according to the guidelines of the World Health Organization[12]. The waist circumference was measured using a flexible, inelastic tape around the natural waistline; the narrowest area between the chest and hips was used as a reference value, as proposed by the World Health Organization[13].

Assessment of Food Consumption

Food consumption was recorded using a three-day food diary comprising two different days during the week and one on the weekend. While submitting forms, guidelines were provided as to the correct way of recording the diet, such as how to list the types of meals, their preparation and portioning, portion sizes, and times at which they were consumed. Upon receipt, the records were checked by researchers. Macronutrients, and calcium and vitamin D content of the diet were calculated using the Dietpro 5.i® software (AS Sistemas, Viçosa, Brazil). To verify the adequacy of the macronutrient intake, acceptable distribution ranges of macronutrients were used as reference; for calcium and vitamin D, the *Estimated Average Requirement* (EAR) was used as a reference value, corresponding to 800 mg/day and 10 μg/day, respectively[14,15].

Determination of Serum and Urinary Cortisol

Venous blood samples of 5 mL were collected from fasting participants in the morning, between 7:30 and 8:30 AM in anticoagulant-free tubes for cortisol assay. Serum was separated from clotted blood by centrifugation at 1831×g for 15 minutes. Assessment of serum cortisol levels was performed by a chemiluminescence method, and the standard morning reference range was taken as 6–28.5 μg/dL.

Urine was collected over a period of 24 hours in demineralized plastic bottles of 5 L capacity with no preservatives, using demineralized funnels that were provided to the study participants securely sealed and labelled with the required instructions. Samples were kept refrigerated until returned to the researcher. The urine was homogenized and the 24-hour volume was measured using a 100 mL graduated cylinder. Then, one aliquot of the collected urine was decanted into a 40 mL plastic vial with no preservatives and stored at –20º C for analysis later. Free cortisol in the urine was determined by a chemiluminescence method, using a standard reference range of 28.5–213.7 μg/24 h.

Statistical Analysis

Data were analysed using the SPSS software version 15.0 for Windows® (SPSS Inc, Chicago, IL, USA). The Kolmogorov-Smirnov test was applied to verify data normality. To compare the outcomes between the two groups, the Student's t-test and Mann-Whitney U-test were used for parametric and non-parametric data, respectively. In addition, the Pearson's coefficient test was used to identify any potential correlations between parametric data sets, and the Spearman's coefficient test was used in non-parametric data sets. The difference was considered statistically significant when the p value was < 0.05, adopting a 95% confidence interval.

RESULTS

Table 1 shows the mean values and standard deviations for age, weight, height, body mass index, and waist circumference, used to assess the nutritional status of morbidly obese patients and control subjects. There was a statistically significant difference in the body weight, body mass index, and waist circumference between the two groups.

Table 1 — Mean values and standard deviations for age, weight, height, body mass index and waist circumference of morbidly obese women and control group

Parameters	Morbidly Obese (n=29) Mean ± SD	Control Group (n=29) Mean ± SD	p
Age (years)	31.8 ± 8.28	28.5 ± 7.15	0.109
Body Weight (kg)	112.98 ±10.43*	56.5 ± 4.88*	<0.001
Height (cm)	1.61 ± 0.55	1.60 ± 0.6	0.558
BMI (Kg/m^2)	43.71 ± 3.18*	22.12 ± 1.57*	<0.001
WC (cm)	114.45 ± 9.08*	71.76 ± 4.02*	<0.001

BMI = Body Mass Index; WC = Waist Circumference; SD = standard deviation; p = p value.
*Difference statistically significant between morbidly obese and control groups, Student's t-test ($p < 0.05$).

The mean values and standard deviations for energy consumption and amount of macronutrients found in the diets consumed by morbidly obese and normal-weight individuals are shown in Table 2. There was no significant difference in the energy and macronutrient intake between the groups ($p > 0.05$).

Table 2 — Mean values and standard deviations for energy consumption and amount of macronutrients of morbidly obese women and control group

Energy/Nutrients	Morbidly Obese (n=29) Mean ± SD	Control Group (n=29) Mean ± SD	p
Energy consumption (kcal)	1814.92 ± 472.09	1920.64 ± 568.70	0.444
Carbohydrate (%)	46.63 ± 6.82	46.32 ± 5.71	0.850
Protein (%)	20.55 ± 3.94	21.05 ± 6.67	0.730
Lipid (%)	32.86 ± 5.82	33.89 ± 5.41	0.485

Student's t-test ($p > 0.05$). SD=standard deviation; p=p value.
Reference values: 10%–35% for protein, 20%–35% for lipids, and 45%–65% for carbohydrates (IOM 2005).

Table 3 shows the mean values and standard deviations of dietary intake of calcium and vitamin D in the morbidly obese and control groups. There was a significant difference in the consumption of these micronutrients ($p < 0.05$) between the groups, and morbidly obese women had a lower intake of calcium and vitamin D.

Table 3 — Mean values and standard deviations of dietary intake of calcium and vitamin D in morbidly obese and control group

Parameters	Morbidly Obese (n=29) Mean ± SD	Control Group (n=29) Mean ± SD	p
Calcium (mg)	761.54 ± 353.21	1004.02 ± 543.27	0.049*
Vitamin D (μg)	8.07 ± 9.86	42.92 ± 56.18	0.002**

*Difference statistically significant between morbidly obese and control groups, Student's t-test ($p < 0.05$).**Mann-Whitney U-test ($p < 0.05$); SD = standard deviation; p = p value.
Reference values: Calcium: EAR=800 mg/day (19-70 years); Vitamin D: EAR=10 μg/day (IOM 2011).

The correlation analysis between the dietary calcium and vitamin D intake and adiposity parameters is shown in Table 4. The correlation between calcium intake and adiposity parameters was not significant ($p > 0.05$); however, there was a significant correlation between vitamin D intake and waist circumference and body mass index ($p < 0.05$).

Table 4 — Correlation analysis between the dietary calcium and vitamin D intake and adiposity parameters

Parameters	Calcium		Vitamin D	
	r	p	r	p
WC	–0.186	0.162	–0.483	< 0.001*
BMI	–0.147	0.271	–0.468	< 0.001*

BMI = Body Mass Index; WC = Waist Circumference; r = correlation coefficient; p = p value.
*Spearman's coefficient test ($p < 0.05$).

Mean values and standard deviation values for serum and urinary cortisol levels of morbidly obese and control subjects are provided in Table 5. No statistically significant difference was found between groups regarding these parameters ($p > 0.05$).

Table 5 — Mean values and standard deviation values for serum and urinary cortisol levels of morbidly obese and control subjects

Parameters	Morbidly Obese (n=29) Mean ± SD	Control Group (n=29) Mean ± SD	p
Serum Cortisol (μg/dL)	9.81 ± 5.75	8.16 ± 4.37	0.260
Urinary Cortisol (μg/dL)	104.98 ± 121.89	104.84 ± 44.24	0.995

Mann-Whitney U-test ($p > 0.05$). Reference values: Serum Cortisol (6–28.5 μg/dL); Urinary Cortisol (28.5–213.7 μg/24h). SD = standard deviation; p = p value.

Table 6 shows the results for correlation analysis between the dietary calcium and vitamin D intake and cortisol concentrations in serum and urine in morbidly obese women. The correlation between these parameters was not significant ($p > 0.05$).

Table 6 — Correlation analysis between the dietary calcium and vitamin D intake and cortisol concentrations in serum and urine in morbidly obese women

Parameters	Serum Cortisol (μg/dL)		Urinary Cortisol (μg/dL)	
	r	p	r	p
Calcium (mg/day)	0.116	0.387	0.005	0.978
Vitamin D (μg/day)	–0.171	0.199	–0.181	0.347

Pearson's coefficient test ($p > 0.05$); r = correlation coefficient; p = p value.

DISCUSSION

This study estimated the amount of dietary calcium and vitamin D intake and investigated the correlation between dietary intake of these micronutrients and cortisol metabolism in morbidly obese women.

Morbidly obese women evaluated in this study ingested calcium content below the EAR values, the difference being statistically significant when compared to the control group. These results can be explained by the fact that the control group had higher intake of food rich in calcium, such as milk and its derivatives. Similar results were verified by Jones et al.[5] who found deficiency in the calcium intake of obese patients.

The present study also observed a higher intake of vitamin D for the control group when compared to morbidly obese women. This result can be attributed to increased consumption of vitamin D food sources for the control group, such as fish of cold and deep water, and foods such as sushi and sashimi. It is important to mention that these food sources of vitamin D were not part of the diet of the morbidly obese evaluated in this study.

It is noteworthy that reduced intake of vitamin D plays an important role in the pathogenesis of obesity, contributing to the increase and accumulation of visceral fat and altering calcium absorption[16]. However, one cannot conclude that obese women in this study have vitamin D deficiency, since this vitamin can also be acquired by exposure to sunlight.

It is worth noting that low levels of calcium in the diet seem to favour the accumulation of body fat. In this regard, Suliburska et al.[17] identified a significant association between central adiposity and low calcium intake in obese women. However, the present study found no significant correlation between dietary calcium intake and the adiposity parameters evaluated.

On the other hand, we found a negative correlation between adiposity parameters (body mass index and waist circumference) and vitamin D intake, emphasizing the important role of this vitamin in maintaining proper body weight. Similar results were obtained by Marcotorchino et al.[18] and Wamberg et al.[10].

Vitamin D deficiency seems to favour the accumulation of body fat, and this process may be linked to the deposit of this vitamin in adipocytes, reducing its bioavailability and causing a cascade of reactions by the hypothalamus that results in increased sensation of hunger and decreased energy expenditure. This situation also increases levels of parathyroid hormone, reduces insulin sensitivity, and increases the concentration of intracellular calcium disproportionately[9].

In relation to average concentrations of serum and urinary cortisol evaluated in this study, participants showed values within the normal range with no statistically significant difference between groups. These results are consistent with those of Rask et al.[19] and Martins et al.[20] who also found that cortisol levels were similar in obese individuals when compared to subjects with normal weight.

An important consideration in this discussion is the dysfunction in cortisol metabolism in visceral obesity. Thus, increasing the metabolic clearance and the hyper-responsiveness of the hypothalamic-pituitary-adrenocortical axis may contribute to the maintenance of adequate levels of this hormone in serum[20,21].

The results of this study did not show the presence of biochemical hypercortisolism in morbidly obese patients. However, it is worth mentioning that the presence of functional hypercortisolaemia present in adipose tissue may contribute to metabolic syndrome by stimulating the production of this hormone in adipose tissue and liver[22].

Correlation analysis shows no significant result between the content of calcium and vitamin D in the diet and serum and urinary cortisol concentrations in both groups. This result can be explained in terms of the limitations of the study by the small number of patients evaluated and the lack of estimation of serum levels of these nutrients, as Zemel and Sobhani[23] found a relationship between the suppression of vitamin D serum levels and the reduction in cortisol production in adipose tissue.

According to Zemel and Sobhani[23] it is possible to state the relationship between vitamin D and the production of cortisol, by stimulating the action of 11β-HSD-1 enzyme that participates in the synthesis of this hormone in human adipocytes. Moreover, Morris and Zemel[24] showed that diets rich in calcium suppress the vitamin D serum levels, reducing cortisol production by adipocytes.

Considering the complexity of the mechanisms involved in obesity, the authenticity of human test data, and disagreement in the literature data on this subject, more research is needed to explore the effects of micronutrients in obesity, which can certainly contribute to a better understanding of its influence on the metabolism of various hormones, such as cortisol.

CONCLUSION

Morbidly obese women evaluated in this study had reduced intake of calcium and vitamin D in amounts below the EAR, without manifesting the influence of these micronutrients on concentrations of serum and urinary cortisol. The vitamin D intake was negatively correlated with adiposity parameters, which demonstrates the involvement of this vitamin in preventing obesity. In this study, the morbidly obese women did not have abnormal concentrations of serum and urinary cortisol; thus, hypercortisolaemia and hypercortisoluria were not seen in the participants.

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